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Wiggling Fingers at the Force-Length Relationship:
First Dorsal Interosseous, Architectural Influence on Force Potential.

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Abstract

Understanding human muscle function is important for many aspects related to the biomechanics community. Developments like microprocessor prosthetics and brain–machine interface for stroke rehabilitation require compete understanding of human muscle function. This understating of muscle function manifests as the musculoskeletal model and provide researchers a means to integrate technology with human function. Despite the advancement in the musculoskeletal model over the last decade, little is known about how human muscle produces sub-maximum force. Moreover, there is no standard means to quantify sub-maximum muscle function. This is due to the majority of research investigating muscle function solely under maximum activation despite the fact that normal human function is primarily a function of sub-maximum conditions. Therefore, the purpose of this study is to investigate sub-maximum properties of the force-length relationship by comparing two methods identified by de Fontana and Herzog (2016).

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Introduction

Skeletal muscle is responsible for producing movement in the human body like an engine is responsible for motion in a vehicle. As bundles of skeletal muscle cells (muscle fascicles) contract, actin proteins slide past myosin proteins, causing the muscle to become shorter (Huxley, 1957). Connective tissue (tendon) attaches opposing ends of skeletal muscle to bones that are connected by a joint; motion is produced as the whole muscle becomes shorter and pulls on the skeleton, decreasing the angle between respective bones.

When a motor vehicle increases force, however, the transmission changes the ratio of the car's speed with respect to the engine speed. This ability to change gears is what allows the car to operate effectively in different conditions; the engine can reduce speed for higher vehicle speeds (favoring velocity) and increase speed for slower vehicle speeds (favoring force). Modeling a vehicle's performance allows researchers to simulate experiments that would be expensive and dangerous. This style of research increases the quality of life for many people by insuring safe and efficient transportation. In skeletal muscle, force production has been modeled for many years (Jones & Round, 1990). Musculoskeletal modeling is a means to quantify and ultimately comprehend muscle function, which allows researchers to performed experiments on computers that otherwise, would be impracticable to take place in a living organism. Mathematical explanation of muscle function is critical for many fields of research such as rehabilitation of muscle injury, treatment of muscular disease, and health or fitness training. Therefor the relationships underlining muscle function must be quantified for the improvement of the musculoskeletal model.

The relationship between force production and the length of muscle has been an important component for understanding muscle function for quite some time. One of the first depictions of the force-length relationship used dissected semitendinosus muscle fibers of frogs to show that maximum isometric force is produced at a specific fiber length (Ramsey & Street, 1940). Moreover, the graphical representation of this relationship, known as the force-length curve, produced an inverted 'U' shape. The vertex of this force-length curve, representing the fiber length that produced the maximum isometric force, was identified by the authors as the resting length of a fiber. Properties of maximum isometric force were not the only parameters of force defined by theses pioneers of the force-length relationship; "the term resting tension refers here to that tension exhibited by unstimulated single muscle fibers when subjected to passive stretches…" (p.17). The importance of resting tension was found within the connective tissue that surrounds a muscle fiber, the sarcolemma. One of the arguments for connective tissue properties being responsible for resting tension was supported by showing that the thickness of a fiber was not indicative of the magnitude of maximum isometric force: "… there may be rather wide variation in thickness that are not related to the size of the fiber." (p.19). Furthermore, a fiber length that produced high force would not have an equally large resting tension. A third property of force quantified within this study was the rate at which force was produced. The relationship can be seen within the slope of the force-length curve. Figure 1 is a reprint of Figure 3 (Ramsey & Street, 1939; p21). The rate of tension development is greater for fiber lengths below resting length (<100 on the x axes)

Figure 1: Force-length curve for a single fiber.

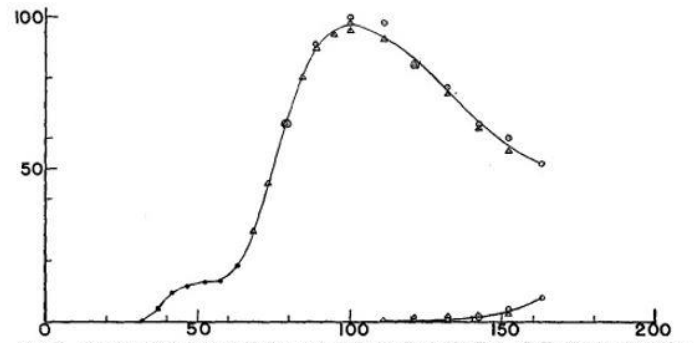


Fig. 3 Length—total tension diagram of an isolated muscle fiber. Ordinate: total tension in per cent of maximum at resting length. Abscissa: lengths in per cent of the resting length (100). Open circles: tensions recorded in ascending order of lengths; triangles: return in descending order; solid circles: irreversible range. Upper curve: active muscle; lower curve: resting muscle.

Source: Ramsey & Street (1939)

compared to the slower rate of development for lengths greater than the resting length (>100 on the x axes). The limitations of this study are well-defined by the authors, reporting that the influential source of error for the force-length curve was in the measurement of the fiber length which was assumed to be 5%.

Until recently, the force-length relationship has been evaluated exclusively with isolated muscle under maximum activation despite the fact that maximum activation is atypical of human function. For example, the previously defined resting length of a muscle fiber is similar to a commonly used definition of optimum muscle length: "the length at which a muscle can produce maximum isometric force" (Holt & Azizi, 2014, p.1). Up to this point, resting length and optimum muscle length are exclusively quantified with static muscle in a state of maximum activation.

The observation made previously by Ramsey and Street (1939) regarding the rate of muscle tension development was extended beyond isometric force leading to the quantification of force-velocity properties. In the study by Zuurbier and Huijing (1992), five gastrocnemius medialis muscles from rats were used to show the speed of

total muscle shortening at optimum length was be explained by the speed of fascicle (84%) and connective tissue (8%) shortening. This relationship was found to vary as optimum length changes. Below optimum muscle length, fascicle and connective tissue shortening were 34 percent and 31 percent of total muscle shortening speed, respectfully. The intent of the authors was to evaluate an assumption of a fixed tendon made in previous literature by mathematically modeling the elastic properties of the tendon. Isokinetic contractions at 80, 85, 90, 95, and 100 percent optimum muscle length were measured along a fixed distance with a transducer attached to the tendon of the rat muscles. The muscle fascicles' angle to the tendon (pennation angle) was measured by triangulating the gastrocnemius medialis with three metal markers. Figure 2 is a reprint of figure 1 (Zuurbier & Huijing, 1992; p1018).

Figure 2: Pennation Angle Measurement of the Gastrocnemius Medialis

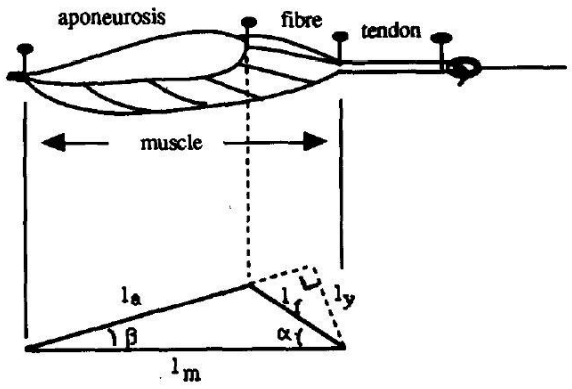


Fig. 1. Schematic representation of the GM muscle–tendon complex, markers inserted and geometrical representation of the muscle. The lengths of elements measured by cinematographic images are indicated: fibre length (l_f), aponeurosis length (l_a), muscle length (l_m), perpendicular distance between the aponeuroses (l_y), fibre angle (α) and aponeurosis angle (β).

Source: Zuurbier & Huijing (1992)

Markers were placed on the proximal and distal tendon and the "distal end of the distal fiber bundle (muscle belly)" (p.1018). The angle of the hypotenuse to the short

leg was assumed to represent the pennation angle. This study was one of the first of its kind and alluded to a variable relation between fascicle speed of shortening and fascicle length without changing the speed of muscle contraction. Furthermore, investigation of the assumption of a fixed tendon was extended to constant aponeuroses length. The authors found support for constant muscle volume while presenting evidence that the length of aponeuroses sheaths vary with muscle length, allowing them to incorporate aponeuroses changes into the musculoskeletal model. "These changes, given the model condition of constant longitudinal section area, should, therefore, be accompanied by some change of perpendicular distance between the aponeuroses, for which experimental indication was found" (p.1023). This study was limited in part by the use of non-human muscle tissue and by the two-dimensional genomic approach used to estimate pennation angle. A right triangle was formed using the general shape of the muscle, assuming that the internal parameters of muscle architecture, specifically the fascicles themselves, was represented by the general size and shape of the entire muscle. There is evidence that the internal arrangement of fascicles is complex and rotates in three dimensions within four first dorsal interosseous (FDI) from two cadavers (Infantolino & Challis, 2012).

The investigation of muscle function has frequently required inference on structures and systems unseen from observations of a larger structure. As described above, Zuurbier and Huijing (1992) observed external shape changes of a muscle to measure internal behavior of fascicles. Though this is not always an ideal approach for muscle modeling, it often is required. It is advantageous to investigate all possible perspectives of muscle function seeking to understand the limits hidden within each.

Azizi, Brainerd, and Thomas (2008) continued to quantify shape and pennation angle variability for a musculoskeletal model. The study investigated a turkey's lateral gastrocnemius, building on the prior work of rats' gastrocnemius medialis muscles (Zuurbier & Huijing, 1992). The authors hypothesized that the change of a muscle's shape was indicative of pennation changes. If the angle at which force was acting increases, the muscle fascicles must rotate, and the muscle was predicted to increase in muscle thickness. Muscle thickness was defined as the distance between two connective tissue sheaths of a muscle, the superficial and deep aponeuroses. This claim would challenge any assumption of constant muscle thickness that has been made in past literature, and present a relationship between thickness and force of a muscle. As fascicles rotate, the overall muscle velocity will exceed the velocity of the fascicle, therefore altering the "muscle's architectural gear ratio (AGR)" (p.1745). If the pennation of the muscle remains constant, the fascicles would not rotate, and the overall muscle velocity will not exceed the velocity of the fascicle. Muscle shape, fascicle velocity, whole muscle velocity, and pennation angle were measured during a series of isotonic contractions of an in situ turkey lateral gastrocnemius. The contractions ranged from 10 to 100 percent isometric forces while initial fascicle length, pennation angle, and total fascicle strain were consistent. Force of each condition was constant during each measurement to control for elastic tissue properties. A significant negative relationship was found between muscle thickness and muscle force (no reported correlation coefficient), supporting the research hypothesis that changes in force acting on a muscle change the behavior of muscle fascicles and ultimately the AGR of the muscle. This relationship between force and

fascicle rotation is similar to the vehicle engine and transition analogy. As force acting on a muscle decreases, the thickness of a muscle will increase along with increases in pennation angle which is the rotation of the fascicles. Just as the transition of a vehicle will favor velocity by changing the gear ratio and slowing the engine for higher vehicle speeds, this study reported a 40 percent higher muscle velocity compared to fascicle velocity for low-force contraction. Alternatively, as force acting on a muscle increases, the thickness of a muscle will decrease along with a similar decrease in pennation angle which is not associated with the rotation of the fascicles, and the reported muscle velocity will equal the fascicle velocity. The analogy follows that a transition will allow the engine to operate faster at lower vehicle speeds. Therefore, pennation of pennate muscle was not constant as previously believed, but was shown to vary as force acted upon it. This study was limited by the use of only a single turkey's lateral gastrocnemius and the use of two-dimensional right triangles for pennation angle measurements. The error accolated with the two-dimensional assumption for fascicles action cannot be quantified until three-dimensional observations can be made within functioning muscle tissue.

This observation of a muscle's ability to vary the speed of fascicle shortening relative to the speed of muscle contraction dependent on the acting forces brought the definitive use of maximal activation into question. As stated earlier, maximal activation is not typical for normal muscle function yet it had been the standard procurer for quantifying the force-length properties. Holt and Azizi (2014) used this same logic to seek an explanation for a previously observed phenomenon that "muscle optimum length increases with decreased activation" (p.1). They claimed

that absolute force would explain the shift in optimum muscle length, not calcium sensitivity. They studied a bullfrog plantaris muscle in vitro during isometric contractions for different levels of activation, "maximal tetanic (high force, high calcium), submaximal tetanic (low force, high calcium) and twitch (low force, low calcium)" (p.1). It was concluded that absolute force explained the increase in optimum muscle length because the submaximal tetanic activation that produced 40 percent peak force (low force, high calcium) was not significantly different in percent optimum muscle length compared to the percent optimum muscle length of the twitch activation (low force, low calcium) levels. A major limit to this study was that the relationship between the stimulation condition and calcium concentrations was not quantified but assumed on the basic understanding of muscle physiology. Also, the relationship between sub-maximum force and optimum muscle length was dependent on the "… force produced by the muscle relative to its maximum force output…" (p.1). It is worth noting that maximum force and optimum muscle length are defined in this study when a muscle is in a state of maximum activation, and some investigators believe that force and activation are not linear (de Fontana & Herzog, 2016).

de Fontana and Herzog (2016) outright refuted the broadly accepted definition for optimum muscle length, "the length at which a muscle can produce maximum isometric force" (Holt & Azizi, 2014, p.1); They stated that "by definition" (p.1268), optimum length confines peak force to a joint angle (muscle tendon unit length) of a muscle group regardless of muscle activation; therefore, classifying sub-maximum force as percentages of absolute force must result in the same joint angle for smaller

percentages of force. An alternative definition for optimum fascial length was proposed: “the length at which the activation-or corresponding energy cost (Hug et. al. 2004)-required to produce a specific force is minimum…” (p.1273). With a new perspective of optimum fascial length, the levels of sub-maximum force may be defined for percentages of absolute activation. In order to compare the force-defined optimum fascial length against the activation-defined optimum fascial length, two maximum isometric knee extensor contractions were performed for 10 joint angles between 80° and 170°. Knee extensor torque, joint angles, muscle activation, and fascicle length were recorded for the vastus lateralis (VL) of nine participants. Figure 3 is a reprint of figures 3 and 4 (p.1271-1272) where definitions of optimum muscle length were compared with force-length curves for 10 levels of force ranging from 10 percent to 100 percent (colored curves). Both force length graphs have peak force for sub-maximum force levels plotted with a black ‘X’. The graph on the left shows peak force for decreasing percentages of absolute force occur at the same joint angle (100 degrees) and longer optimum fascicle lengths. The curve on the right, defining

Figure 3: Force length curves for separate definitions of optimum muscle length.

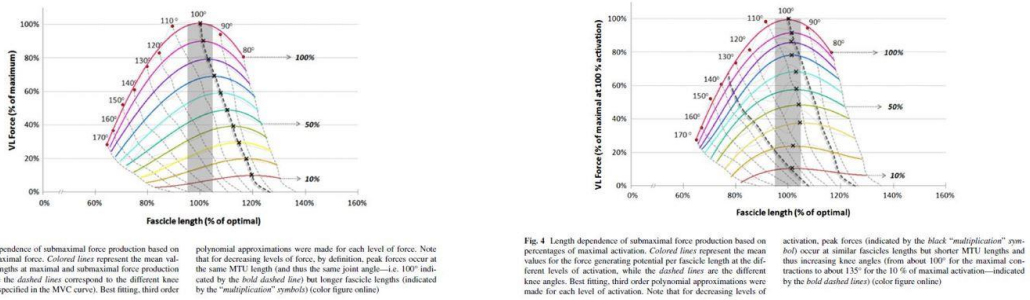


Fig. 3 Length dependence of submaximal force production based on percentages of maximal force. *Colored lines* represent the mean values for fascicle lengths at maximal and submaximal force production (0–100 %), while the *dashed lines* correspond to the different knee angles analyzed (specified in the MVC curve). Best fitting, third order polynomial approximations were made for each level of force. Note that for decreasing levels of force, by definition, peak forces occur at the same MTU length (and thus the same joint angle—i.e. 100° indicated by the *bold dashed line*) but longer fascicle lengths (indicated by the "*multiplication*" *symbols*) (color figure online)

Fig. 4 Length dependence of submaximal force production based on percentages of maximal activation. *Colored lines* represent the mean values for the force generating potential per fascicle length at the different levels of activation, while the *dashed lines* are the different knee angles. Best fitting, third order polynomial approximations were made for each level of activation. Note that for decreasing levels of activation, peak forces (indicated by the *black* "*multiplication*" *symbol*) occur at similar fascicles lengths but shorter MTU lengths and thus increasing knee angles (from about 100° for the maximal contractions to about 135° for the 10 % of maximal activation—indicated by the *bold dashed lines*) (color figure online)

Source: de Fontana & Herzog (2016)

optimum muscle length by maximum force produced with minimum activation, plots the sub-maximum force at percent intervals of activation, resulting in peak force for

decreasing percentages of absolute activation occurring at decreasing joint angles and similar fascicle lengths. The authors found that activation and force were not linear, causing force-based optimum muscle length to vary with decreasing sub-maximum muscle function. In order to keep a consistent optimum fascial length that represents optimum contractile protein overlay, sub-maximum force must account for decreasing percentages of activation. "At a submaximal force level of 10 %, the average fascicle length at peak force was 23% longer than the optimal fascicle length at maximum force ($p < 0.001$)" (p.1271). Some limitations for this study were that the physiological cross-sectional area was used to estimate force contribution from the vastus lateralis as a fraction of all muscles acting over the knee joint, fascicles and aponeuroses sheets were assumed straight, and the patellar tendon moment arm was predicted by a regression equation.

Normal muscle function typically produces sub-maximum force with sub-maximum muscle activation. However, the relationship between force and activation have been identified as non-linear. Therefore, it is important to understand the implications and limits between controversial definitions of sub-maximum muscle function. The force-length relationship during sub-maximum muscle function depends weather muscle function is perceived as a percentage of absolute force or activation. Musculoskeletal models quantify muscle force as a measure of muscle function and are important tools for rehabilitation research. Muscle activation is the mechanism of motor control; understanding how healthy muscle mediate force at all levels of muscle activation can improve and optimize rehabilitation for musculoskeletal injuries or the cognitively impaired. The differences between sub-

maximum activation and sub-maximum force have been quantified *in vivo* using the vastus lateralis, which required an assumption for force discrimination of a system of muscles acting over the knee. Therefore, the purpose of this thesis is to quantify and compare the differences between estimated optimum fascicle length using sub-maximum force and sub-maximum activation with the first dorsal interosseous in order to improve the musculoskeletal model.

Hypotheses

H_o: Repeated manual digitization of ultrasound images of the first dorsal interosseous is not significantly different in optimum fascicle length at 20, 40, 60, 80, and 100% percent sub-maximum muscle function approaches between two investigators.

H_1: Repeated manual digitization of ultrasound images of the first dorsal interosseous is significantly different in optimum fascicle length at 20, 40, 60, 80, and 100% percent sub-maximum muscle function approaches between two investigators.

H_o: First dorsal interosseous optimum fascicle length will not occur at significantly different fascicle lengths for different sub-maximal muscle functions.

H_1: First dorsal interosseous optimum fascicle length will occur at significantly different fascicle lengths for different sub-maximal muscle functions.

Operational Definitions

Aponeurosis: "Aponeuroses are fibrous membranes, of a pearly white color, iridescent, and glistening, which represent very much flattened tendons. They consist of closely packed, parallel, collagenous bundles, and by this characteristic may be differentiated from the fibrous membranes of fascia which have their collagenous bundles more irregularly interwoven. They are only sparingly supplied with blood

vessels" (Goss, 1959, p.306). The superficial and deep aponeurosis is connected by muscle fascicles that shorten to produce muscle force.

Fascicle: A bundle of muscle fibers wrapped in connective tissue (Jones & Round, 1990).

Fascicle Length: Fascicle length calculated according to de Fontana & Herzog (2016); the muscle thickness divided by the sine of the pennation angle. The length of a fascicle is represented by the length of a right triangle's hypotenuse and assumed straight in 2 dimensions.

First Dorsal Interosseous (FDI): The FDI is first, and largest, of four muscles between the dorsal side of the metacarpal bones with a bipennate arrangement of two heads. The lateral head originates from the base of the first metacarpal bone and the medial head originates from the second metacarpal bone. The triangle-shaped muscle is inserted to the lateral portion of the index finger by a tendon. The FDI acts on the metacarpophalangeal joint for abduction of the index finger and adduction of the thumb at the carpometacarpal joint. (Goss, 1959).

Force-length Curve: A graphical representation of the force a muscle can produce at various fascicle lengths (Ramsey & Street, 1940).

Isometric Force: "The force at zero velocity of shortening" (Jones & Round, 1990, p.27).

Muscle Activation: The voltage amplitude on the surface of a muscle that is influenced by the motor cortex. The root mean square (RMS) of EMG voltage amplitude is calculated in windows of 100nm (de Fontana & Herzog, 2016) throughout the five second ramping contraction with the fallowing formula.

$$RMS = \left(\frac{1}{S}\sum_{1}^{S} f^2(s)\right)^{\frac{1}{2}}$$

Where S is the window width and f(s) is the EMG data (Delsys Bagnoli, Delsys Inc. Boston, MA, USA). The EMG signal was then normalized with the maximum RMS value for each joint angle.

Muscle Length: The total length between tendon attachment on opposing ends of the muscle. The length of the whole muscle may also be referred to as the muscle tendon unit length and is dependent of joint angle at which it acts.

Muscle Moment Arm: The length change in a muscle's tendon divided by the joint angle.

Muscle Thickness: The distance between the superficial and deep aponeuroses measured from ultrasound imaging.

Optimum Fascicle Length: The dependent variable and defined: "where the force is greatest for a given amount of activation or energy expenditure" (de Fontana & Herzog, 2016, p.1273). Optimum fascicle lengths at maximal and submaximal forces and activations were predicted by the force-length equation from Otten (1987).

Optimum Muscle Length: The length of a muscle when the whole muscle produces maximum isometric force. "The length at which a muscle can produce maximum isometric force is optimum length" (Holt & Azizi, 2014, p.1).

Pennation Angle: The angle of a muscle fascicle to the deep aponeurosis.

Pennate Muscle: Muscle which "the fibers insert into the tendons at acute angles" (Jones & Round, 1990, p.108).

Sub-Maximum Muscle Conditions: The two independent variables are percent muscle force and percent muscle activation. Each independent variable has five levels; 20, 40, 60, 80, and 100 percent of maximum muscle force and percent of muscle activation respectfully.

Tendon Length: Length of connective tissue connected to either end of the muscle belly.

Limitations

This study may be limited by measurement accuracy of the architectural properties of the First Dorsal Interosseous. Architectural properties are measured through ultrasound image analysis with implementation of open source MATLAB program (Infantolino, 2013). Pennation angle, and muscle thickness are average measurements for all identified fascicles in each ultrasound frame. The algorithm's ability to identify and track fascicles may be limited by the quality of ultrasound imaging, the user-defined region of interest, the contrast gain value, the orientation value, a minimum fascicle size tolerance, and the line-like quality setting. Fascicles are assumed straight in a two-dimensional plane and calculated by muscle thickness and pennation angle measurements. The implications for fascicle rotation in three dimensions have been investigated (Holt & Azizi, 2014). The quality of ultrasound imaging and EMG may be limited by the small volume of the first dorsal interosseous. Muscle moment arm measurement may be limited by the identification of muscle boundaries on the ultrasound images. This study may also be limited by sample size.

Delimitations

The results are only generalized to the right First Dorsal Interosseous. External validity for this study is limited by only one method of measurement for each parameter. Therefore, the results may only compare to studies that implement similar measurement methods.

Method

Participants

This study evaluated the right FDI of four live male participants. Participants were free of any past musculo-skeletal injuries relating to their right hand. Four volunteers, the subject matter expert, one kinesiology major, and two mechanical engineers conducted the skeletal muscle architecture measurements on the same ultrasound images after receiving a period of instruction on the associated physiological relationship between the FDI fascicle pennation angle and ultrasound imagining to evaluate investigator bias of the measurement methods. The subject matter expert was also asked to conduct four repeated measurements on the same ultrasound images to further evaluate the methods reliability. The participants and investigators read and signed an informed consent form approved by the University Institutional Review Board prior to collecting and evaluating subject specific data.

Instruments

EMG system (SP-WO2A-1652, Bagnoli, Delsys Inc. Boston, MA, USA). The EMG system recorded surface EMG signal with the EMGWorks (Delsys Bagnoli, Delsys Inc. Boston, MA, USA) data acquisition software and a 16 channel Desktop System (Bagnoli-16 SEMG System).

EMG sensor (Trigno™ Mini, Bagnoli, Delsys Inc. Boston, MA, USA). The electrode was placed on the medial portion of the FDI medial head adjacent to the ultrasound probe. Identifying motor activation in the FDI with surface EMG has been

shown to have a 95% accuracy when validated against the intramuscular EMG (iEMG) method (Hu, Rymer, & Suresh, 2014).

Alcohol prep pad (REF6818, Webcol, Covidien, Mansfield, MA, USA). A 1in x 1in alcohol wipe was used to clean the area of skin in contact with the EMG electrode to remove any lotion, oils, and dead skin cells.

Ultrasound gel pad (REF04-02, Aquaflex, Parker Laboratories, Fairfield, NJ, USA). A 2cm x 9cm gel pad was cut in half and placed on top of the FDI medial head adjacent to the EMG electrode for enhanced quality of ultrasound imaging.

Ultrasound system (128 CEXT-1Z, Echo Blaster, Telemed Medical Systems, lithuania, Italy). In vivo ultrasound videos were recorded of the FDI for data processing with MATLAB with the EchoWave II software (Version 4.2). The validity and reliability of muscle architectural properties have been assessed in a systematic review (Kwah, Pinto, Diong, & Herbert, 2013) of 42 independent studies resulting in interclass correlation coefficient (ICC) and correlation coefficient (r values) greater than 0.6, and coefficient of variation values less than 10 percent for fascicle length measurement reliability; ICC and r values were greater than 0.5, and coefficient of variation values were less than 14 percent for pennation angle measurements reliability.

Ultrasound probe (HL9.0/60/128Z). The ultrasound probe was placed in the longitudinal plain of the FDI medial head and adjusted for optimum image quality focusing on the distal attachment of the transverse (superficial) head.

Load cell (SSMF-AJ-1000, Interface, Scottsdale, AZ). Used to measure force production of the MCP joint.

MATLAB (R2016b). Costume MATLAB code was used to filter and process the EMG and force data, and identify the associated ultrasound frames at 20, 40, 60, 80, and 100 percent occurrences of force and activation. A second costume MATLAB code was used to manually measure muscle thickness, approximate pennation angle, and calculate fascicle length from each associated ultrasound frame. A third costume MATLAB code created by Dr. Infantolino implemented the force-length equation from Otten (1987) to model the parameters for force-length curves at maximal and submaximal forces and activations.

Metronome (MR800, Matrix). To ensure accuracy of timed phases during each ramping contraction.

Graphing calculator **(TI-84 Plus, Texas Instruments, Dallas, TX, USA).** The random number generator function was used to randomize the order of conditions for each participant.

Procedures

The participants signed a University's Institutional Review Board-approved informed consent before data collection procedures began. A copy was provided for their records.

A Delsys trigger module was uses with a costume two tailed mouse to integrate the Echo Blaster ultrasound system with the force transducer and EMG system. Each participant was seated upright with their right arm and shoulder supported by the table throughout the data collection process to decrees fatigue. A wooden board with angle measurement of 0, 5, 10, 15, and 20 degree markings was used to hold the second metacarpophalangeal (MCP) joint in the desired positions

while the first, third, fourth, and fifth MCP joint were fixed. A force transducer was clamped to the board at the distal end of the lateral side of the second finger and connected directly to the EMG system by a wireless transmitter. An EMG sensor was attached to the web space between the first and second fingers after the skin's surface was thoroughly cleaned with an alcohol and brillo pad. An ultrasound gel pad (Aquaflex) was cut in half to form a semi-circle and the straight edge was placed adjacent to the EMG sensor. The ultrasound probe was placed on top of the gel pad, longitudinally to the medial FDI head. Ultrasound images of the right FDI were taken at each of the five degree intervals of the second MCP joint abduction and processed for muscle moment arm measurement at a later date. Two contractions were conducted for 0, 5, 10, 15, and 20 degree MCP joint angles while EMG amplitude and ultrasound videos were recorded. Contractions followed a ramping protocol resembling the methods adapted by de Fontana and Herzog (2016); ramping from 0 to 100% isometric force over a three second interval, and holding 100% isometric force for two additional seconds. A Matrix metronome (MR800) was used along with the EMG Acquisition software's (version 4.2) that provided visual and auditory feedback encouraging the adherence of each ramping contraction. The trial with acceptable ultrasound video quality and highest total force for each joint angle was used for analysis. The order of joint angle trials was randomized with the random number generator function from a Texas Instruments calculator (TI-84 Plus). A minimum of two-minute rest period was strictly enforced between each contraction. On average each participant completed the testing procedure within 45 minutes.

Design and Analysis

The independent variable for this design was sub-maximum muscle conditions of the FDI, percent muscle force and percent muscle activation. Each independent variable has five levels; 20, 40, 60, 80, and 100 percent of maximum muscle force and percent of muscle activation respectfully. The dependent variable was optimum fascicle length predicted by the force-length equation from Otten (1987).

The first measure was the proportion of force at 20, 40, 60, and 80 percent maximum isometric force. The second measure was the proportion of force at 20, 40, 60, and 80 percent maximum muscle activation.

Costume MATLAB code was used to process and filter all data. Ten data arrays were read into MATLAB, five containing force and five containing EMG activation data for each joint angle ramping condition trial. The ultrasound video and force graph was displayed for both ramping protocols at each of the five joint angles. The trial with an acceptable ultrasound video quality and highest absolute force was selected for processing and the remaining data arrays and ultrasound videos were removed. EMG voltage data was processed in the following order, 20Hz high-pass Butterworth filter, full-wave rectification, smoothed by a 1 Hz low-pass Butterworth filter, and normalized with respect to maximum activation of each ramp. Then the force voltage data was processed in the following order, 1 Hz low-pass filter, the magnitude of the data was taken, and the data was normalized with respect to maximum force of each ramp. Figure 4 is an example of a processing figure output from one subject at zero degrees MCP flection.

Figure 4: Stages of Data processing for EMG and Force

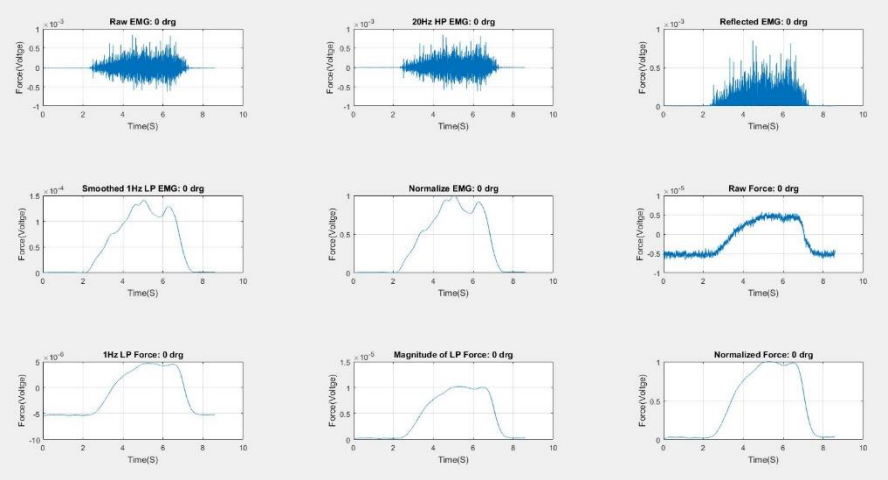


Figure 4 is one example of a 3x3-matrix plot created by the custom MATLAB data processing code to document each stage in the filtering of EMG and force data.

After the force and activation data was processed the felted data was saved and all raw and unused variables were removed. The sub-maximum levels of activation and force were found and saved to respective excel files with the time of occurrences.

A second MATLAB source file was used to analysis the ultrasound images of the FDI using sub-maximum occurrences output saved from the data processing file. Each ultrasound video was played and paused at the associated frame of each sub-activation and sub-force percentage. The investigator was than required to select six points on each ultrasound image. The first two points measured the muscle thickness by identifying the superficial and deep aponeuroses in the middle of the muscle belly near the end of the visible portion of the deep aponeuroses. The two points were saved on the ultrasound frame as a blue line. The fallowing two points (sections 3 and 4) were then made such that they form a red line on the distal end of the transverse head's superficial aponeuroses. The investigators were instructed to choose a region that was as long as possible without representing aponeuroses curvature. The angle of

this red line was taken to be the arctangent of the absolute value of its slope. The final two points (selections 5 and 6) were used to mark the deep aponeuroses angle at the distal portion in similar fashion as the superficial aponeuroses. The pennation angle was assumed to be represented by the sum of the superficial and deep aponeuroses two angles of the. The fascicle length was calculated by dividing the muscle thickness by the sine of the pennation angle. Figure 5 shows an example of an ultrasound frame saved by the source code after the measurements were taken.

Figure 5: Ultrasound frame for 0 degrees MCP flection at 20% activation.

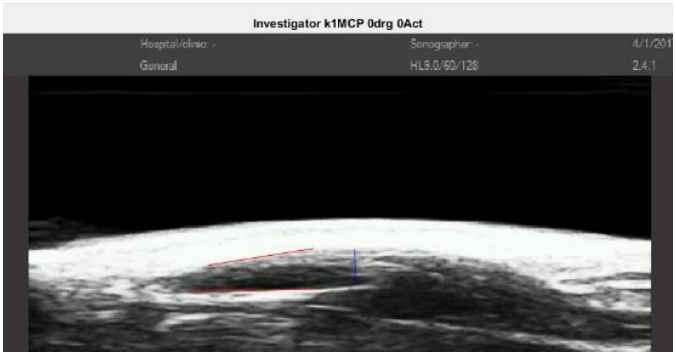


Figure 4 is one example of the six points that the investigator was required to selected in order to measure the architecture properties of the first dorsal interosseous. The muscle thickness measurement is represented by the blue line while the pennation angle was calculated with the slope of both red lines. The length of the fascicle was calculated with the resulting thickness and angle measurement.

A data set was saved for every participant at every with the time, normal EMG, normal force, magnitude of force, fascicle length, pennation angle, and muscle thickness for every sub-maximum muscle condition at each joint angle.

The third source code was created by Dr. Infantolino and used the analyzed ultrasound data with the force-length equation from Otten (1987) to model the parameters for force-length curves and output fascicle length as a percentage of optimum length.

Results

Reliability Tests

Results of the independent t-test suggest that there is a significant difference in mean optimum fascicle length measurements at 20% activation based sub-maximum muscle function between the subject-matter expert (M = 95.34, SD = 8.13) and the primary investigator (M = 118.85, SD = 3.30), $t(6) = -5.60, p < 0.05$. Bonferroni correction was applied for ten comparisons resulting in non-significance, $t(3) = -5.60, p > 0.005$. No significant differences were found between the subject-matter expert and the primary investigator's repeated measurements of optimum fascicle length at 40% activation, $t(6) = 0.281, p > 0.05$, 60% activation, $t(6) = 0.06, p > 0.05$, 80% activation, $t(6) = 0.39, p > 0.05$, 100% activation, $t(6) = 0.33, p > 0.05$, 20% force, $t(6) = -0.09, p > 0.05$, 40% force, $t(6) = -1.47, p > 0.05$, 60% force, $t(6) = -1.30, p > 0.05$, 80% force, $t(6) = -0.59, p > 0.05$, 100% force, $t(6) = 0.33, p > 0.05$. Table 1 presents the means and standard deviations from two investigator's repeated measure of the same data.

Table 1

Means and Standard Deviations of Optimum Fascicle Length

	The Subject-Matter Expert			
	Sub-Maximum Activation		Sub-Maximum Force	
Percentages	M	SD	M	SD
20%	95.34	8.13	107.46	9.01
40%	110.12	12.64	104.43	6.85
60%	97.99	17.16	105.55	5.51
80%	94.46	19.49	110.26	8.35
100%	105.5	21	105.5	10.5
	The Primary Investigator			
20%	118.58	3.3	108.31	8.58
40%	100.74	15.13	115.02	7.16
60%	97.42	16.93	113.56	8.67
80%	90.39	16.13	104.93	10.58
100%	101.84	10.11	101.84	10.11

*Independent t-test of difference *, $p < 0.005$(Bonferroni correction for ten comparisons)*

Optimum Fascicle Length Significance Test

No significant difference was found in optimum fascicle length at 20% sub-maximal muscle function between the activation and force approaches; $t(4) = -1.10$, $p > 0.05$. No significant difference was found in optimum fascicle length at 40% sub-maximal muscle function between the activation and force approaches; $t(4) = 0.03$, $p > 0.05$. No significant difference was found in optimum fascicle length at 60% sub-maximal muscle function between the activation and force approaches; $t(4) = -2.21$, $p > 0.05$. No significant difference was found in optimum fascicle length at 80% sub-maximal muscle function between the activation and force approaches; $t(4) = -0.78$, $p > 0.05$. No significant difference was found in optimum fascicle length at 100% sub-maximal muscle function between the activation and force approaches; $t(4) = -1.48$, $p > 0.05$. Table 2 presents the means, standard deviations, and mean differences with confidence intervals for each subject calculated by the subject matter expert.

Table 2

Means and Standard Deviations of Different Sub-Maximal Muscle Conditions

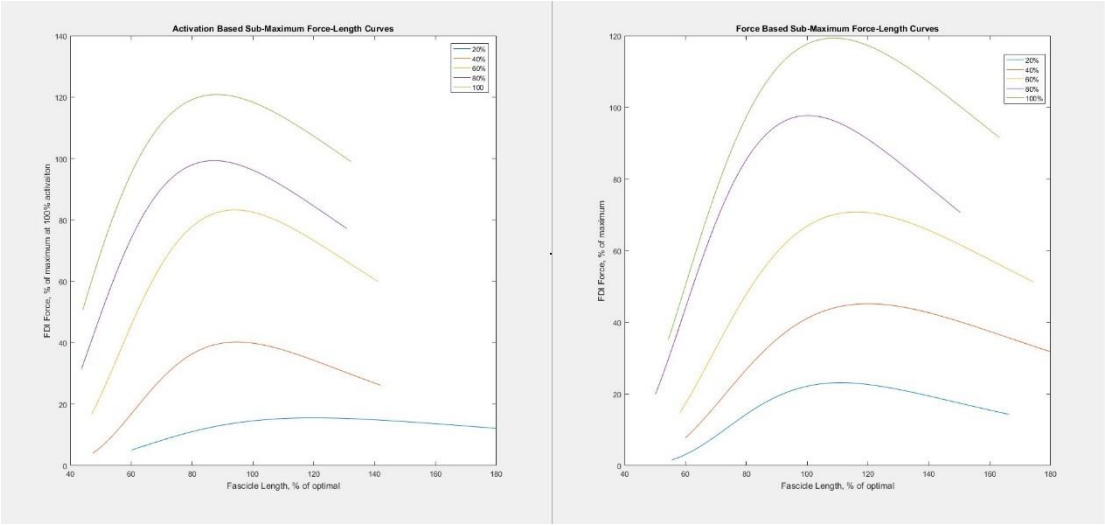
	Sub-Maximum Activation		Sub-Maximum Force	
Percentages	M	SD	M	SD
20%	99.16	15.39	109.96	12.26
40%	101.23	16.31	100.97	10.26
60%	79.44	4.92	99.76	17.75
80%	94.90	22.96	105.54	14.67
100%	94.39	21.12	111.49	9.34
Difference:		M		CI
20%		-10.81		(-36.10, 14.48)
40%		0.26		(-24.50, 25.03)
60%		-20.32		(-49.63, 8.99)
80%		-10.6		(-45.7, 24.4)
100%		-17.1		(-49.2, 15.0)

*Independent t-test of difference *, $p < 0.01$(Bonferroni correction for five comparisons)*

Discussion

The purpose of this study was to compare optimum fascicle length with two perspectives of sub-maximum muscle function. de Fontana and Herzog (2016) found significant differences in optimum fascicle length at sub-maximal muscle force compared to sub-maximal muscle activation in the vastus lateralis of nine subjects. Adapting similar methods and proceeds from the proceeding reference, sub-maximum force-length curves of the first dorsal interosseous were created by incrementing percentages of force and activation separately. Figure 6 is graphical representations of the fitted sub-maximum force-length properties by single subject analysis. However, this study failed to reject the null hypothesis: H_o: First dorsal interosseous optimum fascicle length will not occur at significantly different fascicle lengths for different sub-maximal muscle functions.

Figure 6: Force-Length Curves of the First Dorsal Interosseous at Sub-Maximum and Maximum Muscle Functions



Comparing Figure 6 with Figure 3 provides a visualization for the non-significance found in the present study compared to the significance in longer fascial lengths for decreasing force-based sub-maximum muscle function.

One major challenge to replicating the results of de Fontana and Herzog (2016) was the relative size of the muscle being investigated. The first dorsal interosseous was investigated in order to eliminate the physiological cross-sectional area assumption for force discrimination that is required for any muscle that does not act over a joint independently. The elimination of this assumption provided assurance that all measured force at the second MCP joint was produced by the FDI, unlike the vastus lateralis whitch only contributes a fraction of the force measured over the knee. The vastus lateralis is a larger muscle that provides clear ultrasound reflections of fascicles that can be easily identified and measured. The pennation angles of the FDI were not really measured, requiring a generalization similar to those of Zuurbier and Huijing (1992). Due to the small muscle belly, reflections of the muscle fascicle were unclear and discontinuous. The general shape of the FDI's surrounding connective tissue was assumed to represent the average pennation angle for any given ultrasound frame (see Figure 5). The first hypothesis of this study was written to maintain accountability of this assumption; H_o: Repeated manual digitization of ultrasound images of the first dorsal interosseous is not significantly different in optimum fascicle length at 20, 40, 60, 80, and 100% percent sub-maximum muscle function approaches between two investigators. The failure to reject this null hypothesis implies that the manual digitization of ultrasounds images are relatively repeatable. However, the 20% activation approach for sub-maximum muscle function was significantly different before the Bonferroni correction for 10 comparisons was made. This is an example of the uncertainty that still remains within the pennation angle

measurements of s of the FDI. Further data collection and repeated analyzation is necessary before the methods used in this study can be considered reliable.

When evaluating force-length curves for the FDI one must understand a fundamental printable for muscle function. The FDI, like many muscles, only operates on a small portion of the ascending force-length curve. Again, due to the relatively small muscle and low range of motion (about 0 to 20 degrees) the majority of the force length curve and corresponding optimum fascicle length must be predicted. This was accomplished according to the force-length equation from Otten (1987). Perhaps a muscle such as the vastus lateralis that has a larger range of motion (170 to 80 degrees) will operate on a larger percentage of the force-length curve and not require such extrapolation to produce the force-length curves.

In order to model any physical relationship, reliable methods of measurement must be used. If the measurements are not reliable or reputable, no conclusions can be made about the results. Therefore, a fundamental purpose for this thesis must exist: to test the reliability of repeated muscle architectural measurements made by manual digitization of ultrasound images of the FDI. Both associated null hypothesizes for this objective were not rejected in support of repeatability in the manual digitization of ultrasound images of the first dorsal interosseous for sub-maximum muscle function optimum fascicle length measures.

The methods and analysis presented in this thesis have shown some statistical potential; however, the sample sizes evaluated across the entire study are inadequate to account for the limits of manual digitization, force-length curve extrapolation, and two-dimensional reduction of a three-dimensional relationship. In order to make any

generalizable conclusion more data must be collected with an emphasis on single subject analysis to further establish the validity of this design.

In conclusion, more understanding and standardizations are needed for sub-maximum muscle function; not only for the force-length properties such as the work by de Fontana and Herzog (2016), but for other physiological properties of muscle like force-velocity curves. Moreover, innovated assessment like the work of Holt and Azizi (2014) is required to challenge the 2-dimentenal assumptions for pennation angles and fascicles behavior in order to improve the Musculoskeletal and further the quantification of muscle function.

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Appendices

Data Sets

Data set 1: Subject-Matter Expert

Lfopt_A_60	Lfopt_A_80	Lfopt_A_100	Lfopt_F_20	Lfopt_F_40	Lfopt_F_60	Lfopt_F_80	Lfopt_F_100
95.88	120.00	110.41	115.29	118.90	114.76	120.00	110.41
120.00	99.33	117.10	120.00	105.28	115.15	85.41	117.10
97.95	80.29	120.00	113.82	107.63	93.83	115.65	120.00
78.12	78.23	74.56	80.72	85.92	98.46	120.00	74.56

Fascicle length as a percentage of optimum length calculated by repeated manual digitization of ultrasound images of the first dorsal interosseous and modeled by force-length equation from Otten (1987). All measures of a single subject.

Data set 2: Primary Investigator

Lfopt_A_20	Lfopt_A_40	Lfopt_A_60	Lfopt_A_80	Lfopt_A_100	Lfopt_F_20	Lfopt_F_40	Lfopt_F_60	Lfopt_F_80	Lfopt_F_100
120.00	82.05	76.06	63.78	117.28	91.21	116.14	120.00	102.91	117.28
117.34	114.51	116.03	109.47	90.14	102.77	117.05	120.00	108.23	90.14
120.00	110.19	95.00	99.37	93.29	104.34	120.00	103.32	82.62	93.29
120.00	85.79	72.31	69.99	113.14	116.61	120.00	109.92	103.10	113.14
109.51	105.73	120.00	84.73	87.77	110.18	111.63	120.00	98.45	87.77
118.92	120.00	105.04	114.12	105.53	115.24	120.00	116.80	113.40	105.53
120.00	119.86	102.11	98.13	95.01	120.00	118.32	95.41	97.74	95.01
120.00	82.32	78.56	80.04	99.78	105.08	97.54	110.19	120.00	99.78
120.00	92.17	97.70	93.58	108.01	114.10	119.90	120.00	114.16	108.01
120.00	94.80	111.42	90.68	108.45	103.61	109.60	120.00	108.68	108.45

Fascicle length as a percentage of optimum length calculated by repeated manual digitization of ultrasound images of the first dorsal interosseous and modeled by force-length equation from Otten (1987). All measures of a single subject.

Data set 3: Subject data

	Lfopt_A_20	Lfopt_A_40	Lfopt_A_60	Lfopt_A_80	Lfopt_A_100	Lfopt_F_20	Lfopt_F_40	Lfopt_F_60	Lfopt_F_80	Lfopt_F_100
n1	92.35	120.00	86.69	103.10	88.63	93.88	94.61	120.00	99.36	98.28
n3	84.04	82.15	76.52	120.00	99.51	106.94	112.78	105.42	114.89	120.00
n5	120.00	95.00	76.24	65.41	69.43	120.00	90.46	77.65	87.90	115.22
n8	100.23	107.78	78.30	91.09	120.00	119.03	106.01	95.96	120.00	112.47

Fascicle length as a percentage of optimum length calculated by repeated manual digitization of ultrasound images of the first dorsal interosseous and modeled by force-length equation from Otten (1987). All measures of different subjects.

CONSENT FOR RESEARCH

The Pennsylvania State University

Title of Project: Muscle Activity During Common Lifting Activities

Principal Investigator: Benjamin W. Infantolino, Ph.D.

Address: 114A Beaver Community Center

Telephone Number: 610-396-6153

Subject's Printed Name: _____

We are asking you to be in a research study. This form gives you information about the research.

Whether or not you take part is up to you. You can choose not to take part You can agree to take part and later change your mind* Your decision will not be held against you.

Please ask questions about anything that is unclear to you and take your time to make your choice.

1. **Why is this research study being done?**

We are asking you to be in this research because you are healthy and do not have any injuries to your limbs in the last 18 months. This research is being done to measure activity of your muscles during lifting exercisesr These measurements are important for our understanding of which muscles are active during different variations of the same activity. Understanding this will help researchers to determine the most effective exercises to train or rehabilitate specific muscles.

Approximately 60 people will take part in this research study at Penn State Berks

2. **What will happen in this research study?**

- Once you arrive at the Kinesiology Lab the procedures will be explained to you and once all your questions have been satisfactorily answered and you are willing to participate you will be asked to sign the informed consent form.
- Electromyography (EMG) leads will be attached to the subject with hypoallergenic tape over the muscles of interest to measure the electrical activity of the muscles.
- Hypoallergenic gel will be placed on your skin to facilitate clear images for the ultrasound equipment. The ultrasound probe will be held on your skin using light pressure.
- Participants will perform three trials while voluntarily moving their joint through a normal range of motion for each lifting activity. Joint motion will not be

restricted in any way. The velocity of joint motion will be controlled through the use of a metronome.

- Steps 2 and 3 will be repeated for each muscle and lifting activity of interest.
- Once all images are taken, the researcher will remove the electrodes and you will be done with the study, no follow-up is required.

3. **What are the risks and possible discomforts from being in this research study?**
   There is a risk of loss of confidentiality if your information or your identity is obtained by someone other than the investigators, but precautions will be taken to prevent this from happening. The only discomfort you would experience would be when the electrodes are removed (i.e. taking off a bandage) or local fatigue from using a specific muscle. Both risks are minimal and if the subject experiences discomfort the researcher will work with the subject to ensure that discomfort is minimized or eliminated.

4. **What are the possible benefits from being in this research study?**
   The potential benefits of this study include a deeper understanding of the function muscles during movement. This increased understanding can translate to improved rehabilitative care for musculoskeletal disorders or injury.

5. **What other options are available instead of being in this research study?** You may decide not to participate in this research.

6. **How long will you take part in this research study?**
   If you agree to take part, it will take you about 2 hours to complete this research study, You will be not be asked to return to the research site.

7. **How will your privacy and confidentiality be protected if you decide to take part in this research study?**
   Efforts will be made to limit the use and sharing of your personal research information to people who have a need to review this information.

   Your research records will be labeled with an identifying number (i.e. subject 1) and will be kept in a locked file cabinet and on a password protected network drive.

   In the event of any publication or presentation resulting from the research, no personally identifiable information will be shared.

   We will do our best to keep your participation in this research study confidential to the extent permitted by law. However, it is possible that other people may find out about your participation in this research study. For example, the following people/groups may check and copy records about this research.

- The Office for Human Research Protections in the U. S. Department of Health and Human Services
- The Institutional Review Board (a committee that reviews and approves research studies) and
- The Office for Research Protections.

Some of these records could contain information that personally identifies you. Reasonable efforts will be made to keep the personal information in your research record private. However, absolute confidentiality cannot be guaranteed.

8. What happens if you are injured as a result of taking part in this research study?

In the unlikely event you become injured as a result of your participation in this study, medical care is available. It is the policy of this institution to provide neither financial compensation nor free medical treatment for research-related injury. By signing this document, you are not waiving any rights that you have against The Pennsylvania State University for injury resulting from negligence of the University or its investigators.

9. What are your rights if you take part in this research study?

Taking part in this research study is voluntary.

- You do not have to be in this research.
- If you choose to be in this research, you have the right to stop at any time.
- If you decide not to be in this research or if you decide to stop at a later date, there will be no penalty or loss of benefits to which you are entitled.

12. If you have questions or concerns about this research study, whom should you call?

Please call the head of the research study (principal investigator), Benjamin Infantolino at 610-396-6153 if you:

- Have questions, complaints or concerns about the research.
- Believe you may have been harmed by being in the research study.

You may also contact the Office for Research Protections at (814) 865-1775, ORProtections@psu.edu if you:

- Have questions regarding your rights as a person in a research study.
- Have concerns or general questions about the research.
- You may also call this number if you cannot reach the research team or wish to talk to someone else about any concerns related to the research.

INFORMED CONSENT TO TAKE PART IN RESEARCH

Signature of Person Obtaining Informed Consent

Your signature below means that you have explained the research to the subject or subject representative and have answered any questions he/she has about the research.

_____	_____	_____	_____
Signature of person who explained this research	Date	Time	Printed Name

(Only approved investigators for this research may explain the research and obtain informed consent.)

Signature of Person Giving Informed Consent

Before making the decision about being in this research you should have:

- Discussed this research study with an investigator,
- Read the information in this form, and
- Had the opportunity to ask any questions you may have.

Your signature below means that you have received this information, have asked the questions you currently have about the research and those questions have been answered. You will receive a copy of the signed and dated form to keep for future reference.

Signature of Subject

By signing this consent form, you indicate that you voluntarily choose to be in this research and agree to allow your information to be used and shared as described above.

_____	_____	_____	_____
Signature of Subject	Date	Time	Printed Name